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5. A method of producing a mammal having a predetermined sex as described in claim 1 wherein said female of said species of said mammal has uterine horns and wherein said step of inserting said sorted artificial insemination sample into said female of said species of said mammal comprises the step of inserting said sorted artificial insemination sample both ipsi- and contra-lateral within the uterine horns of said female of said species of said mammal.

6. A method of producing a mammal having a predetermined sex as described in claim 1 or 2 wherein said female of said species of said mammal has at least one uterine horn and wherein said step of inserting said sorted insemination sample into said female of said species of said mammal comprises the step of inserting said sorted artificial insemination sample deep within said uterine horn.

7. A method of producing a mammal having a predetermined sex as described in claim 5 wherein said step of inserting said sorted artificial insemination sample into a female of said species of said mammal further comprises the step of inserting said sorted artificial insemination sample deep within said uterine horns.

8. A method of producing a mammal having a predetermined sex as described in claim 6 wherein said step of inserting said sorted artificial insemination sample into a female of said species of said mammal further comprises the step of inserting said sorted artificial insemination sample within said uterine horn through the use of embryo transfer equipment.

9. A method of producing a mammal having a predetermined sex as described in claim 7 wherein said step of inserting said sorted insemination sample into a female of said

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or 3 wherein said steps of determining the sex characteristic of a plurality of said sperm cells and sorting said sperm cells according to the determination of their sex characteristic comprise the steps of:

- a. establishing a cell source which supplies sperm cells to be sorted;
- b. chemically coordinating a sheath fluid to create a sheath fluid environment for said sperm cells which is coordinated with both a pre-sort and a post-sort sperm cell fluid environment;
- c. sensing a property of said sperm cells;
- d. discriminating between said sperm cells having a desired sex characteristic; and
- e. collecting said sperm cells having the desired sex characteristic.

15. A method of producing a mammal having a predetermined sex as described in claim 1 wherein said steps of determining the sex characteristic of a plurality of said sperm cells and sorting said sperm cells according to the determination of their sex characteristic comprise the steps of:

- a. establishing a cell source which supplies bovine sperm cells to be sorted;
- b. establishing a sheath fluid for said bovine sperm cells which contains about 2.9% sodium citrate;
- c. sensing a property of said bovine sperm cells;
- d. discriminating between said bovine sperm cells having a desired sex characteristic; and
- e. collecting said bovine sperm cells having the desired sex characteristic.

16. A method of producing a mammal having a predetermined sex as described in claim 1 wherein said steps of determining the sex characteristic of a plurality of said sperm cells and sorting said sperm cells according to the determination of their sex characteristic comprise the steps of:

- a. establishing a cell source which supplies equine sperm cells to be sorted;
- b. establishing a sheath fluid for said equine sperm cells which contains a hepes buffered medium;
- c. sensing a property of said equine sperm cells;
- d. discriminating between said equine sperm cells having a desired sex

collecting said equine sperm cells having the desired sex characteristic.

e. collecting said equine sperm cells having the desired sex characteristic.

17. A method of producing a mammal having a predetermined sex as described in claim 1 wherein said steps of determining the sex characteristic of a plurality of said sperm cells and sorting said sperm cells according to the determination of their sex characteristic comprise the steps of:

a. establishing a cell source which supplies sperm cells to be sorted;

b. establishing a sheath fluid for said sperm cells;

c. sensing a property of said sperm cells;

10      d.      discriminating between said sperm cells having a desired sex characteristic; and  
e.      collecting said sperm cells having the desired sex characteristic while cushioning  
said sperm cells from impact with a collector wherein a cushioning element  
comprises initial collection fluid in the bottom of said collector and wherein said  
collector has a configuration sufficiently large to avoid impact of said sperm cells  
15      with said collector.

with said collector.

18. A method of producing a mammal having a predetermined sex as described in claim 1 wherein said steps of determining the sex characteristic of a plurality of said sperm cells and sorting said sperm cells according to the determination of their sex characteristic comprise the steps of:

a. establishing a cell source which supplies sperm cells to be sorted;

b. establishing a sheath fluid for said sperm cells;

c. sensing a property of said sperm cells;

d. discriminating between said sperm cells having a desired sex characteristic; and

e. collecting said sperm cells having the desired sex characteristic in a citrate collection fluid containing about six percent egg yolk prior to commencing said step of collecting.

19. A method of producing a mammal of a desired sex as described in claim 15, 16, 17 or 18 and further comprising the step of sorting said sperm cells at a rate of at least 500 sorts per second.

$\frac{1}{\sqrt{\pi}} \int_{-\infty}^{\infty} f(x) e^{-x^2} dx = \frac{1}{\sqrt{\pi}} \int_{-\infty}^{\infty} f(x) e^{-x^2} dx$

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30. An improved flow cytometer system for isolating sperm cells for producing a mammal according to the method of claim 1 comprising:

- a. a cell source which supplies sperm cells to be analyzed by the flow cytometer;
- b. a sheath fluid source which creates a sheath fluid environment for said sperm cells;
- c. a nozzle through which said sperm cells pass while subjected to said sheath fluid environment;
- d. an oscillator which acts upon said sheath fluid as it passes through said nozzle;
- e. a sperm cell sensing system which responds to said sperm cells;
- f. a sorter discrimination system which acts to sort said sperm cells according to the determination of their sex characteristic at a rate of at least 500 sorts per second; and
- g. a collector into which sorted sperm cells having a desired sex characteristic are placed.

15 31. An improved flow cytometer system for isolating sperm cells for producing a mammal as described in claim 30 wherein said sheath fluid source which creates a sheath fluid environment for said sperm cells contains citrate buffer.

32. An improved flow cytometer system for isolating sperm cells for producing a mammal as described in claim 30 wherein said sheath fluid source which creates a sheath fluid environment for said sperm cells contains a hepes buffered medium.

33. An improved flow cytometer system for isolating sperm cells for producing a mammal as described in claim 32 wherein said cell source comprises equine sperm cells.

34. An improved flow cytometer system for isolating sperm cells for producing a mammal as described in claim 31 wherein said cell source comprises bovine cells.

25 35. An improved flow cytometer system for isolating sperm cells for producing a mammal as described in claim 34, wherein said sheath fluid source contains 2.9% sodium citrate buffer.



36. An improved flow cytometer system for isolating sperm cells for producing a mammal as described in claim 30, 31, 32, 33, 34 or 35 wherein said collector is used to provide a sorted artificial insemination sample having from about 10% to about 50% of the number of said sperm cells relative to the typical unsorted artificial insemination sample.

10 38. An improved flow cytometer system for isolating sperm cells for producing a mammal  
as described in claim 37 wherein said configuration sufficiently large to avoid impact of  
said sperm cells with said collector comprises a collection container having a diameter  
of at least fifteen millimeters.

- a. collecting bovine sperm cells from a male of a species of bovine mammal;
- b. determining the sex characteristic of a plurality of said bovine sperm cells;
- c. sorting said bovine sperm cells according to the determination of their sex characteristic, wherein a sheath fluid environment for said bovine sperm cells contains about 2.9% sodium citrate;

25 e. inserting said artificial insemination sample into a female of said species of said bovine mammal;

g. producing an offspring bovine mammal of the desired sex.

40. A method of producing a mammal having a predetermined sex as described in claim 39 wherein said step of fertilizing at least one egg within said female of said species of said bovine mammal comprises the step of fertilizing at least one egg within said female species of said bovine mammal at success levels of at least 90%.
- 5 41. A method of producing a mammal having a predetermined sex as described in claim 39 or 40 wherein said steps of inserting said bovine sorted artificial insemination sample into a female of said species of said bovine mammal and fertilizing at least one egg within said female of said species of said bovine mammal at success levels of at least 50% is accomplished in a field environment.
- 10 42. A method of producing a mammal having a predetermined sex as described in claim 39 wherein said steps of inserting said bovine sorted artificial insemination sample into said female of said species of said bovine mammal and fertilizing at least one egg within said female of said species of said bovine mammal at success levels of at least 50% in a field environment comprises the steps of repetitively inserting a significant number of bovine sorted artificial insemination samples into a significant number of said females of said species of said bovine mammal in rapid succession and in farm or ranch conditions.
- 15 43. A method of producing a mammal having a predetermined sex as described in claim 39, 40 or 41 wherein said bovine mammal has uterine horns and wherein said step of inserting said sorted bovine artificial insemination sample into said female species of said bovine mammal comprises the step of inserting said bovine sorted artificial insemination sample both ipsi- and contra-lateral within the uterine horns of said bovine mammal.
- 20 44. A method of producing a mammal having a predetermined sex as described in claim 39, 40 or 41 wherein said bovine mammal has at least one uterine horn and wherein said step of inserting said sorted bovine artificial insemination sample into said female species of said bovine mammal comprises the step of inserting said bovine sorted artificial insemination sample deep within said uterine horn.
- 25 45. A method of producing a mammal having a predetermined sex as described in claim 42



establishing an unfrozen sorted insemination sample, wherein said step of sorting said sperm cells according to the determination of their sex characteristic occurs at a sorting time, and wherein said step of inserting said insemination sample into a female of said species of said mammal occurs not later than about ten hours from said sorting time.

- 5 51. A method of producing a mammal having a predetermined sex as described in claim 1 or 50 wherein said step of determining the sex characteristic of a plurality of said bovine sperm cells comprises the step of staining said bovine sperm cells with at least about 38 micro-molar content of stain.
- 10 52. A method of producing a mammal having a predetermined sex as described in claim 39 wherein said steps of determining the sex characteristic of a plurality of said bovine sperm cells and sorting said bovine sperm cells according to the determination of their sex characteristic, wherein a sheath fluid environment for said bovine sperm cells contains about 2.9% sodium citrate comprise the steps of:
- 15 a. establishing a cell source which supplies bovine sperm cells to be sorted;
- b. chemically coordinating said sheath fluid environment for said bovine sperm cells which is coordinated with both a pre-sort and a post-sort cell fluid environment;
- c. sensing a property of said bovine sperm cells;
- d. discriminating between said bovine sperm cells having a desired sex characteristic; and
- 20 e. collecting bovine sperm cells having said desired sex characteristic.
- 25 53. A method of producing a mammal having a predetermined sex as described in claim 40 wherein said steps of determining the sex characteristic of a plurality of said bovine sperm cells and sorting said bovine sperm cells according to the determination of their sex characteristic, wherein a sheath fluid environment for said bovine sperm cells contains about 2.9% sodium citrate comprise the steps of:
- a. establishing a cell source which supplies bovine sperm cells to be sorted;
- b. chemically coordinating said sheath fluid environment for said bovine sperm cells which is coordinated with both a pre-sort and a post-sort cell fluid environment;
- c. sensing a property of said bovine sperm cells;

d. discriminating between said bovine sperm cells having a desired sex characteristic; and

e. collecting bovine sperm cells having said desired sex characteristic.

- 10 a. establishing a cell source which supplies bovine sperm cells to be sorted;  
b. establishing said sheath fluid environment for said bovine sperm cells;  
c. sensing a property of said bovine sperm cells;  
d. discriminating between said bovine sperm cells having a desired sex  
characteristic; and  
e. collecting said bovine cells having said desired sex characteristic while  
15 cushioning said bovine sperm cells from impact with a collector wherein a  
cushioning element comprises initial collection fluid in the bottom of said  
collector and wherein said collector has a configuration sufficiently large to avoid  
impact of said sperm cells with said collector.

- 25      a.      establishing a cell source which supplies bovine sperm cells to be sorted;  
b.      establishing said sheath fluid environment for said bovine sperm cells;  
c.      sensing a property of said bovine sperm cells;  
d.      discriminating between said bovine sperm cells having a desired sex  
characteristic; and  
e.      collecting said bovine sperm cells having the desired sex characteristic in a citrate  
30      collection fluid containing about six percent egg yolk prior to commencing said

step of collecting.

56. A method of producing a mammal of a desired sex as described in claim 39 and further comprising the step of sorting said bovine sperm cells at a speed of at least 500 sorts per second.
- 5 57. A method of producing a mammal having a predetermined sex as described in claim 39 and further comprising the step of using an ovulatory pharmaceutical to cause multiple eggs to be produced by said female of said species of said bovine mammal and wherein said step of fertilizing at least one egg within said female of said species of said bovine mammal at success levels of at least 50% comprises the step of fertilizing a plurality of  
10 said eggs produced by said female of said species of said bovine mammal to produce multiple, sexed embryos having a predetermined sex.
58. A method of producing a mammal having a predetermined sex as described in claim 57 wherein said step of using an ovulatory pharmaceutical to cause multiple eggs to be produced by said female of said species of said bovine mammal comprises the step of  
15 injecting a dosage of follicle stimulating hormone a plurality of times a day wherein said ovulatory pharmaceutical is injected in half day increments between any of days 2 and 18 of the estrus cycle.
59. A method of producing a mammal having a predetermined sex as described in claim 58 wherein said step of injecting said dosage of follicle stimulating hormone a plurality of  
20 times a day comprises the step of injecting said follicle stimulating hormone in approximately half day increments at a dosage level of 6, 6, 4, 4, 2, 2, 2, and 2 mg between days 9 and 12 inclusive of the estrus cycle and further comprising the step of injecting 25 and 12.5 mg of prostaglandin F-2-alpha on the sixth and seventh dosages, respectively, of said follicle stimulating hormone.
- 25 60. A method of producing a mammal having a predetermined sex as described in claim 56 wherein said step of sorting said cells at a rate of at least 1200 sorts per second comprises operating a flow cytometer in the range of about 5 kilohertz to about 50 kilohertz.

61. A method of producing a mammal having a predetermined sex as described in claim 54 wherein said step of cushioning said bovine sperm cells from impact with said collection container comprises a collection container having stream matched physical characteristics.

5 62. A method of producing a mammal having a predetermined sex as described in claim 54 wherein said step of cushioning said bovine sperm cells from impact with a collection container comprises the step of providing a collection container having a diameter of at least fifteen millimeters.

63. An improved flow cytometer system for isolating bovine sperm cells for producing a mammal according to the method of claim 39 comprising:

10 a. a cell source which supplies bovine sperm cells to be analyzed by said flow cytometer;

b. a sheath fluid source which creates a sheath fluid environment for said bovine sperm cells which contains about 2.9% sodium citrate;

15 c. a nozzle through which said bovine sperm cells pass while subjected to said sheath fluid environment;

d. an oscillator which acts upon said sheath fluid as it passes through said nozzle;

e. a sperm cell sensing system which responds to said bovine sperm cells;

20 f. a sorter discrimination system which acts to sort said bovine sperm cells having a desired sex characteristic; and

g. a collector into which said bovine sperm cells having said desired sex characteristic are placed.

64. An improved flow cytometer system for isolating bovine sperm cells as described in claim 63 wherein said collector is used to provide a bovine sorted artificial insemination sample.

25 65. An improved flow cytometer system for isolating bovine sperm cells as described in claim 63 or 64 wherein said nozzle, said oscillator, said cell sensing system, and said sorter discrimination system are part of said flow cytometer system and wherein said

flow cytometer system comprises a cell sorter which sorts said bovine sperm cells at a rate of at least 500 sorts per second.

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66. An improved flow cytometer system for isolating bovine sperm cells as described in claim 64 wherein said sorted bovine sperm cells are used to provide bovine sperm cells for artificial insemination.
67. An improved flow cytometer system for isolating bovine sperm cells as described in claim 65 wherein said cell sorter sorts said bovine sperm cells to be analyzed at a rate of at least 1200 sorts per second.
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68. An improved flow cytometer system for isolating bovine sperm cells as described in claim 67 wherein said cell sorter operates in the range of about 5 kilohertz to about 50 kilohertz.
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69. An improved flow cytometer system for isolating bovine sperm cells as described in claim 63 wherein said collector comprises an initial collection fluid in the bottom of said collector and wherein said collector has a configuration sufficiently large to avoid impact of said sperm cells with said collector.
70. An improved flow cytometer system for isolating bovine sperm cells as described in claim 69 wherein said collector comprises a collection container having a diameter of at least fifteen millimeters.
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71. An improved flow cytometer system for isolating bovine sperm cells as described in claim 63 wherein said collector comprises a collection container having stream matched physical characteristics.
72. An improved flow cytometer system for isolating bovine sperm cells as described in claim 63 wherein bovine sperm cells are stained with at least 38 micro-molar content of stain.



73. An improved flow cytometer system for isolating bovine sperm cells as described in claim 63 wherein said sheath fluid source which creates a sheath fluid environment for said bovine sperm cells which contains about 2.9% sodium citrate further comprises a sheath fluid environment which is coordinated with said pre-sort and post-sort bovine sperm cell fluid environments.

- a. collecting equine sperm cells from a male of a species of equine mammal;
- b. determining the sex characteristic of a plurality of said equine sperm cells;
- c. sorting said equine sperm cells according to the determination of their sex characteristic, wherein a sheath fluid environment for said equine sperm cells contains hepes buffered medium;

e. inserting said equine sorted insemination sample into a female of said species of said equine mammal;

g. producing an offspring equine mammal of the desired sex.

75. A method of producing a mammal having a predetermined sex as described in claim 74 wherein said steps of inserting said equine sorted artificial insemination sample into a female of said species of said equine mammal and fertilizing at least one egg within said female of said species of said equine mammal at success levels of at least 35% is accomplished in a field environment.

76. A method of producing a mammal having a predetermined sex as described in claim 74 wherein said steps of inserting said equine sorted artificial insemination sample into a female of said species of said equine mammal and fertilizing at least one egg within said female of said species of said equine mammal at success levels of at least 35% in a field

environment comprises the steps of repetitively inserting a significant number of equine sorted insemination samples into a significant number of females of said species of said equine mammal in rapid succession and in farm or ranch conditions.

5 77. A method of producing a mammal having a predetermined sex as described in claim 74 wherein said equine mammal has uterine horns and wherein said step of inserting said equine sorted artificial insemination sample into a female of said species of said equine mammal comprises the step of inserting said equine sorted artificial insemination sample both ipsi- and contra-lateral within the uterine horns of said equine mammal.

10 78. A method of producing a mammal having a predetermined sex as described in claim 74 wherein said equine mammal has at least one uterine horn and wherein said step of inserting said equine sorted artificial insemination sample into a female species of said equine mammal comprises the step of inserting said equine insemination sample deep within said uterine horn.

15 79. A method of producing a mammal having a predetermined sex as described in claim 77 wherein said step of inserting said equine sorted artificial insemination sample into a female species of said equine mammal further comprises the step of inserting said equine sorted artificial insemination sample deep within said uterine horns.

20 80. A method of producing a mammal having a predetermined sex as described in claim 78 wherein step of inserting at least a portion of said equine sorted artificial insemination sample into a female species of said equine mammal further comprises the step of inserting said equine sorted artificial insemination sample within said uterine horn through the use of embryo transfer equipment.

25 81. A method of producing a mammal having a predetermined sex as described in claim 79 wherein step of inserting said equine sorted artificial insemination sample into a female species of said equine mammal further comprises the step of inserting said equine sorted artificial insemination sample within said uterine horn through the use of embryo transfer equipment.

82. A method of producing a mammal having a predetermined sex as described in claim 77 wherein said step of inserting said sorted artificial insemination sample into a female of said species of said mammal comprises the step of inserting said sorted artificial insemination sample twelve hours after the time which is generally regarded as optimal for a single artificial insemination.
83. A method of producing a mammal having a predetermined sex as described in claim 81 wherein said step of establishing an equine sorted artificial insemination sample comprises the step of establishing an unfrozen equine sorted artificial insemination sample, wherein said step of sorting said equine sperm cells according to the determination of their sex characteristic occurs at a sorting time, and wherein said step of inserting said sorted equine artificial insemination sample into a female of said species of said equine mammal occurs not later than about seventeen hours from said sorting time.
84. A method of producing a mammal having a predetermined sex as described in claim 81 wherein said step of establishing an equine sorted artificial insemination sample comprises the step of establishing an unfrozen equine sorted artificial insemination sample, wherein said step of sorting said equine sperm cells according to the determination of their sex characteristic occurs at a sorting time, and wherein said step of inserting said equine artificial insemination sample into a female species of said equine mammal occurs not later than about ten hours from said sorting time.
85. A method of producing a mammal having a predetermined sex as described in claim 75 wherein said step of determining the sex characteristic of a plurality of said equine sperm cells comprises the step of staining said equine sperm cells with at least about 38 micromolar content of stain.
86. A method of producing a mammal having a predetermined sex as described in claim 74 wherein said steps of determining the sex characteristic of a plurality of said equine sperm cells and sorting said equine sperm cells according to the determination of their sex characteristic, wherein a sheath fluid environment for said equine sperm cells

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- 5 e. collecting said equine sperm cells having the desired sex characteristic while cushioning said equine sperm cells from impact with a collector wherein a cushioning element comprises initial collection fluid in the bottom of said collector and wherein said collector has a configuration sufficiently large to avoid impact of said sperm cells with said collector.

89. A method of producing a mammal having a predetermined sex as described in claim 74 wherein said steps of determining the sex characteristic of a plurality of said equine sperm cells and sorting said equine sperm cells according to the determination of their sex characteristic comprise the steps of:

- 10 a. establishing a cell source which supplies equine sperm cells to be sorted;  
b. establishing a sheath fluid for said equine sperm cells;  
c. sensing a property of said equine sperm cells;  
d. discriminating between said equine sperm cells having a desired sex characteristic; and  
15 e. collecting said equine sperm cells having the desired sex characteristic in a citrate collection fluid containing about six percent egg yolk prior to commencing said step of collecting.

90. A method of producing a mammal of a desired sex as described in claim 74 and further comprising the step of sorting said equine sperm cells at a rate of at least 500 sorts per second.  
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91. A method of producing a mammal having a predetermined sex as described in claim 74 and further comprising the step of using an ovulatory pharmaceutical to cause multiple eggs to be produced by said female of said species of said equine mammal comprises the step of injecting a dosage of follicle stimulating hormone a plurality of times a day  
25 wherein said ovulatory pharmaceutical is injected in half day increments between any of days 2 and 18 of the estrus cycle.

92. A method of producing a mammal having a predetermined sex as described in claim 91 wherein injecting said dosage of follicle stimulating hormone a plurality of times a day



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102. An improved flow cytometer system for isolating equine sperm cells for producing a mammal as described in claim 101 wherein said cell sorter operates in a range of about 5 kilohertz to about 50 kilohertz.

103. An improved flow cytometer system for isolating equine sperm cells for producing a mammal as described in claim 99 wherein said collector comprises an initial collection fluid in the bottom of said collector and wherein said collector has a configuration sufficiently large to avoid impact of said sperm cells with said collector.

104. An improved flow cytometer system for isolating equine sperm cells for producing a mammal as described in claim 103 wherein said collector comprises a collection container having a diameter of at least fifteen millimeters.

105. An improved flow cytometer system for isolating sperm cells according to their sex comprising:

- a cell source which supplies sperm cells to be analyzed by the flow cytometer;
- a chemically coordinated sheath fluid source which creates a sheath fluid environment for said sperm cells which is selected to be coordinated with both a pre-sort and a post-sort sperm cell fluid environment;
- a nozzle through which said sperm cells pass while subjected to said sheath fluid environment;
- an oscillator which acts upon said sheath fluid as it passes through said nozzle;
- a sperm cell sensing system which responds to said cells;
- a sorter discrimination system which acts to sort said sperm cells according to their sex; and
- a collector into which said sperm cells sorted according to their sex are placed.

106. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 wherein said collector has a collection fluid, wherein said collection fluid comprises a nutrient which is coordinated to balance the amount of said nutrient after completion of the sorting of said sperm cells.



107. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 which further comprises a collector into which said sperm cells having a desired characteristic are placed and which comprises a citrate collection fluid containing about six percent egg yolk.

5 108. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 wherein said nozzle, oscillator, cell sensing system, and sorter discrimination system are part of a flow cytometer system and wherein said flow cytometer system comprises a cell sorter which sorts said sperm cells at a rate of at least 500 sorts per second.

10 109. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 wherein said chemically coordinated sheath fluid source which creates a sheath fluid environment for said sperm cells which is selected to be coordinated with both a pre-sort and a post-sort sperm cell fluid environment comprises a citrate.

15 110. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 or 109 wherein said cell source comprises bovine sperm cells.

111. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 wherein said cell source comprises equine sperm cells.

112. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 wherein said collector is used to provide a sorted artificial insemination sample.

20 113. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 112 wherein said sorted insemination sample comprises a sorted artificial insemination sample having no more than about ten percent of the typical number of sperm cells provided in a typical unsorted artificial insemination dosage.

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114. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 112 wherein said sorted artificial insemination sample comprises a bovine sorted insemination sample and wherein said bovine sorted artificial insemination sample comprises a dosage of less than about five hundred thousand bovine sperm.

5 115. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 112 wherein said sorted artificial insemination sample comprises a bovine sorted insemination sample and wherein said bovine sorted artificial insemination sample comprises a dosage of less than about three hundred thousand bovine sperm.

10 116. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 112 wherein said sorted artificial insemination sample comprises an equine sorted insemination sample and wherein said equine sorted artificial insemination sample comprises a dosage of less than about ten million equine sperm.

15 117. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 112 wherein said collector is used to provide a sorted artificial insemination sample for artificial insemination.

118. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 wherein said chemically coordinated sheath fluid source comprises a solution containing a hepes buffered medium.

20 119. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 118 wherein said cell source comprises equine sperm cells.

120. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 wherein said chemically coordinated sheath fluid source comprises a solution containing about 2.9% sodium citrate.

25 121. An improved flow cytometer system for isolating sperm cells according to their sex as described in claim 105 wherein said collector has a cushioning element, wherein said

cushioning element comprises initial collection fluid in the bottom of the said collector and wherein said collector has a configuration sufficiently large to avoid impact of said sperm cells with said collector.

122. An improved flow cytometer system for isolating sperm cells according to their sex as  
5 described in claim 121 wherein said collector comprises a container comprising said cushioning element.

123. An improved flow cytometer system for isolating sperm cells as described in claim 122 wherein said container comprises a collection tube.

124. An improved flow cytometer system for isolating sperm cells as described in claim 123  
10 wherein said collection tube is at least about fifteen millimeters wide.

125. A method of sorting sperm cells using a flow cytometer according to claim 108, comprising the steps of:

- a. establishing a cell source which supplies sperm cells to be sorted;
- b. chemically coordinating a sheath fluid to create a sheath fluid environment for  
15 said sperm cells which is coordinated with both a pre-sort and a post-sort cell fluid environment;
- c. sensing a property of said sperm cells;
- d. discriminating between sperm cells according to their sex; and
- e. collecting sperm cells having the desired sex characteristic.

20 126. A method of sorting sperm cells as described in claim 125, wherein said step of chemically coordinating said collector fluid to create an ending collector fluid environment for said sperm cells which is coordinated with a pre-sort fluid environment.

127. A method of sorting sperm cells as described in claim 125 wherein said step of  
25 establishing a cell source comprises the step of establishing a source of bovine sperm cells.

128. A method of sorting sperm cells as described in claim 125 wherein said step of establishing a cell source comprises the step of establishing a source of equine sperm cells.

129. A method of sorting sperm cells as described in claim 127 wherein said step of chemically coordinating a sheath fluid to create a sheath fluid environment for said sperm cells which is coordinated with both a pre-sort and a post-sort cell fluid environment comprises the step of establishing a cell source which supplies bovine sperm cells to be sorted and establishing a sheath fluid which contains about 2.9% sodium citrate.

130. A method of sorting sperm cells as described in claim 128 wherein said step of chemically coordinating a sheath fluid to create a sheath fluid environment for said sperm cells which is coordinated with both a pre-sort and a post-sort cell fluid environment comprises the step of establishing a cell source which supplies equine sperm cells to be sorted and establishing a sheath fluid which contains a hepes buffered medium.

131. A method of sorting sperm cells as described in claim 125 wherein said step of establishing a cell source comprises the step of providing an initial nutrient for said cells and further comprising the step of providing a collection fluid nutrient for said cells and wherein said step of collecting cells having the desired characteristic in a collector fluid comprises the step of balancing said initial nutrient and said collection fluid nutrient after the completion of said step of collecting said cells.

132. A method of sorting sperm cells as described in claim 125 wherein said step of collecting cells having the desired characteristic in a collector fluid comprises the step of establishing a citrate collection fluid containing about six percent egg yolk prior to commencing said step of collecting.

133. A method of sorting sperm cells as described in claim 132 wherein said step of establishing a cell source comprises the step of establishing a source of bovine sperm cells.

134. A method of sorting sperm cells as described in claim 125 and further comprising the step of inseminating a mammal using a sorted insemination sample.

135. A method of sorting sperm cells as described in claim 125 further comprising the step of collecting sperm cells having the desired sex characteristic comprising cushioning said sperm cells from impact with a collector wherein cushioning said sperm cells comprises collecting said sperm cells into an initial collector fluid in the bottom of a collector and wherein said collector has a configuration sufficiently large to avoid impact of said sperm cells with said collector.

136. A method of sorting sperm cells as described in claim 135 wherein said step of collecting said sperm cells into an initial collector fluid in the bottom of a collector and wherein said collector has a configuration sufficiently large to avoid impact of said sperm cells with said collector comprises the step of providing a collection container.

137. A method of sorting sperm cells as described in claim 136 wherein said step of providing a collection container comprises providing a collection container having a diameter of at least 15 millimeters.

138. A method of sorting sperm cells as described in claim 137 and further comprising the step of sorting said cells at a rate of at least 500 sorts per second.

139. A method of sorting sperm cells as described in claim 138 wherein said step of sorting said cells at a rate of at least 500 sorts per second comprises the step of operating a flow cytometer in a range of about 5 kilohertz to about 50 kilohertz.

140. A method of producing a sexed sperm specimen as described in claim 125 and further comprising the step of sorting said cells at a rate of at least 500 sorts per second.

141. An improved flow cytometer system for isolating sperm cells comprising:

- a cell source which supplies sperm cells to be analyzed by the flow cytometer;

b. a sheath fluid source which creates a sheath fluid environment for said sperm cells;

c. a nozzle through which said sperm cells pass while subjected to said sheath fluid environment;

5 d. an oscillator which acts upon said sheath fluid as it passes through said nozzle;

e. a cell sensing system which responds to said sperm cells;

f. a sorter discrimination system which acts to sort sperm cells according to their sex; and

10 g. a collector having a cushioning element wherein said cushioning element comprises initial collection fluid in the bottom of the said collector and wherein said collector has a configuration sufficiently large to avoid impact of said sperm cells with said collector.

142. An improved flow cytometer system for isolating sperm cells as described in claim 141 wherein said collector comprises a container.

15 143. An improved flow cytometer system for isolating sperm cells as described in claim 144 wherein said container comprises a collection tube.

144. An improved flow cytometer system for isolating sperm cells as described in claim 141 wherein said collection tube is at least about fifteen millimeters wide.

20 145. An improved flow cytometer system for isolating sperm cells as described in claim 142 wherein said container comprises a test tube having the physical characteristics of a stream-matched container.

146. A method of sorting sperm cells according to claim 141, comprising the steps of:

25 a. establishing a cell source which supplies sperm cells to be sorted;

b. establishing a sheath fluid to create a sheath fluid environment for said sperm cells;

c. sensing a property of said sperm cells;

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wherein said collector configured to avoid impact between said sperm cells and said collector comprises a stream-matched container.

152. An improved flow cytometer system for isolating sperm cells as described in claim 151 wherein said collector configured to avoid impact between said sperm cells and said collector comprises a collection tube having a diameter sufficiently large to avoid impact  
5 between said sperm cells and the inner walls of said collection tube.
153. An improved flow cytometer system for isolating sperm cells as described in claim 152 wherein said step wherein said collection tube has a diameter of at least 15 millimeters.
154. An improved flow cytometer system for isolating sperm cells as described in claims 150,  
10 154, 155 or 156 wherein said nozzle, said oscillator, said cell sensing system, and said sorter discrimination system are part of a flow cytometer system and wherein said flow cytometer system comprises a cell sorter which sorts said sperm cells at a rate of at least 500 sorts per second.
155. A method of producing multiple, embryos having predetermined sex from a female mammal comprising:  
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- a. creating superovulation in said mammal to create at least two eggs comprising the step of using an ovulatory pharmaceutical to cause multiple eggs to be produced, wherein said ovulatory pharmaceutical is injected in half day increments between any of days 2 and 18 of the estrus cycle;
  - 20 b. determining a sex of a sperm cell of a male mammal;
  - c. sorting according to said sex of said sperm cells;
  - d. inserting said sorted sperm cells into a uterus of said female mammal after an onset of estrus; and
  - 25 e. fertilizing a plurality of said eggs in said uterus to produce multiple, embryos having predetermined sex.
156. A method of producing multiple, embryos having predetermined sex as described in claim 155 wherein said step for injecting said ovulatory pharmaceutical in half day



increments comprises injecting at least seven injections and further comprising the step of incorporating an estrus manipulation system at least on about the sixth and seventh injections.

157. A method of producing multiple, embryos having predetermined sex as described in  
5 claim 156 wherein inserting said sorted sperm cells into said uterus comprises inserting said sperm cells into both uterine horns of said uterus.

158. A method of producing multiple, embryos having predetermined sex as described in claim 157 wherein inserting into both uterine horns comprises inserting said sperm cells approximately between 20 to 24 hours inclusive after said onset of said estrus.

10 159. A method of producing multiple, embryos having predetermined sex as described in claim 155 wherein said step of using an ovulatory pharmaceutical to cause multiple eggs to be produced comprises the step of injecting a dosage of follicle stimulating hormone a plurality of times a day.

160. A method of producing multiple, embryos having predetermined sex as described in  
15 claim 159 wherein said step of creating superovulation in said mammal to create at least two eggs further comprises the step of incorporating an estrus manipulation system comprising the step of supplementing said dosage of follicle stimulant hormone with prostaglandin F-2-alpha.

20 161. A method of producing multiple, embryos having predetermined sex as described in claim 160 wherein injecting said dosage of follicle stimulating hormone a plurality of times a day comprises injecting said follicle stimulating hormone in approximately half day increments at a dosage level of 6, 6, 4, 4, 2, 2, 2, and 2 mg between days 9 and 12 inclusive of the estrus cycle and wherein supplementing said dosage of follicle stimulating hormone with prostaglandin F-2-alpha comprises the step of injecting 25 and  
25 12.5 mg of prostaglandin F-2-alpha on the sixth and seventh dosages, respectively, of said follicle stimulating hormone.

162. A method of producing multiple, embryos having predetermined sex as described in claim 155 and further comprising the steps of:

- a. staining sperm cells of a male mammal using 38 micro-molar stain;
- b. sorting according to said sex of said sperm cells at a rate of at least 500 sorts per second; and
- c. concentrating said sorted sperm cells.

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163. A method of producing multiple, embryos having predetermined sex as described in claim 155 wherein inserting said sorted sperm cells comprises using a sorted insemination sample.

10 164. A method of producing multiple, embryos having predetermined sex as described in claim 162 wherein inserting said sorted sperm cells comprises using a sorted insemination sample.

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